twin pregnancies of sheep that I have so far had the opportunity to examine, a monochorial condition was found, though the fetuses were dizygotic; but the circulation of each fetus was closed. This appears to be the normal condition in sheep; but if the two circulations should anastomose, we should have the conditions that produce a sterile free-martin in cattle. The possibility of their occurrence in sheep is therefore given.

The fertile free-martin in cattle may be due to cases similar to those normal for sheep. Unfortunately when the first two cases of normal cattle free-martins that I have recorded, came under observation I was not yet aware of the significance of the membrane relations, and the circulation was not studied. But I recorded in my notebook in each case that the connecting part of the two halves of the chorion was narrow, and this is significant. In the third case the two chorions were entirely unfused; this case, therefore, constitutes an *experimentum crucis*. The male was 10.4 cm. long; the female 10.2 cm. The reproductive organs of both were entirely normal. The occurrence of the fertile free-martin is therefore satisfactorily explained.

The sterile free-martin enables us to distinguish between the effects of the zygotic sex-determining factor in mammals, and the hormonic sex-differentiating factors. The female is sterilized at the very beginning of sex-differentiation, or before any morphological evidences are apparent, and male hormones circulate in its blood for a long period thereafter. But in spite of this the reproductive system is for the most part of the female type, though greatly reduced. The gonad is the part most affected; so much so that most authors have interpreted it as testis; a gubernaculum of the male type also develops, but no scrotal sacs. The ducts are distinctly of the female type much reduced, and the phallus and mammary glands are definitely female. The general somatic habitus inclines distinctly toward the male side. Male hormones circulating in the blood of an individual zygotically female have a definitely limited influence, even though the action exists from the beginning of morphological sex-differentiation. A detailed study of this problem will be published at a later date.

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A CHEMOTROPIC RESPONSE OF THE HOUSE FLY (MUSCA DOMESTICA L.)[1]

IT is generally conceded that the house fly lays its eggs most frequently in fermenting vegetable substances. Of these, fermenting horse manure is most often chosen, and about cities probably ninety per cent. or more of the house flies are bred from this substance.[2]

Although manure varies considerably, depending upon the food, the age and the health of the horse, it seems to be invariably attractive to female house flies, provided it is moist and not very old. The flies come to the manure primarily to lay their eggs, and although they may obtain some food from it, this is only a secondary object.

These general observations, together with some preliminary studies recently published,[3] led me to believe that the house fly was allured to the manure pile by the odor of some volatile chemical substance which was liberated during the early stages of fermentation. Acting on this hypothesis, I have tested during the past summer the response of the house fly to a number of inorganic and organic compounds which occur as products of fermentation in barnyard manures.

This paper is a preliminary statement of the results of these experiments. A more detailed account will be given in another place.

Trap Experiments with Ammonia and Other Chemical Substances

The following chemical compounds were exposed in glass containers in screen-wire-fly

[1] This work was done in the department of entomology, New Jersey Agricultural College Experiment Station, and is published by permission of Dr. T. J. Headlee, entomologist of that station.

[2] Howard, L. O., "The House Fly—Disease Carrier," New York, 1911, p. 7.

[3] 27th Ann. Rpt. N. J. Agr. College Exp. Station, 1914, pp. 396–399.

traps, 9¾ inches high, and 6 inches in diameter at the base: Ammonium carbonate U. S. P. (contained about 97 per cent. of ammonium acid carbonate and ammonium carbamate), ammonium sulphide solution, ammonium hydroxide, ethyl alcoholic solutions of skatol and indol, ethyl alcohol, acetic, formic, butyric and valerianic acids, hydrogen sulphide solution and carbon dioxide. The traps which volatilized carbon dioxide were equipped with Erlenmeyer flask droppers, which delivered dilute hydrochloric acid a drop at a time on to bits of limestone in the pan of the trap. By this method a small but fairly constant amount of carbon dioxide was evolved throughout a number of hours. A trap was similarly equipped for use in the ammonium hydroxide experiment.

The experiments were performed at a place where flies were always present, but never excessively abundant.

Negative results were obtained in all but the ammonium hydroxide and ammonium carbonate experiments. The results of ten ammonium carbonate trap experiments are summarized below.

Material Used in Each Trap	Number of Traps	Duration of Experiments	Number of Flies Caught			
			Males	Females	Sex Undetermined	Total
Ammonium carbonate, 85–234 gm., and water (50–90 c.c.) or without water.	23	51–220 hrs.	16	186	3	205
Control, with or without water (50–90 c.c.)...	17	51–220 hrs.	5	3	4	12

House flies were attracted to the traps which contained ammonium carbonate. Small amounts of water and carbon dioxide, both constituents of ammonium carbonate, were not sought by flies, and it is concluded that the other constituent, ammonia, was the attracting agent.

The best results were obtained when water was added to the ammonium carbonate, because it prevented the deposit of a powdery layer of the less volatile ammonium acid carbonate which otherwise hindered the escape of ammonia.

The single ammonium hydroxide trap caught three female house flies during twenty-five hours' exposure.

Since the flies caught in the ammonium carbonate traps were largely females (90.7 per cent.), it was desired to know whether ammonia was particularly attractive to females, or whether females were unusually abundant in the vicinity of the experiments. Under ordinary conditions remote from breeding places the proportion of sexes in the house fly is about equal, with a slight excess of females.[4]

Accordingly, traps baited with food materials (milk, sweet soda water), were maintained in the vicinity of the ammonium carbonate experiments from July 21 to July 29. During this time 274 house flies were captured, 45.9 per cent. of which were males, and 54.0 per cent. females. In the same period, the ammonium carbonate traps caught 65 flies, 7.6 per cent. males and 89.2 per cent. females. Ammonia attracted a great preponderance of females.

Oviposition Experiments

Acidulated horse manure, timothy chaff, pine sawdust, and cotton were treated in such a way that they evolved ammonia. They were then exposed in a place frequented by flies, and after a period which varied from 3 to 99 hours in the individual experiments, counts were made of the egg-masses which had been deposited. Two or more eggs, placed together, were considered an egg-mass, but the large majority of clusters contained many more than two eggs. Occasional single eggs were ignored.

Oviposition in Acidulated Horse Manure.—The purpose of this series of experiments was to show whether fresh horse manure which did not volatilize ammonia would still induce the house fly to oviposit, and whether such manure, when again volatilizing ammonia, would attract the female fly. Fresh horse manure was treated with dilute hydrochloric acid so that

[4] Hewitt, "The House Fly (*Musca domestica* L.)," etc., Cambridge, England, 1914, p. 98.

the free ammonia was converted into ammonium chloride, an involatile salt at the ordinary temperature. The manure was left in a slightly acid state so that all the ammonia formed during the course of the experiments would immediately unite with the acid. It was tested with litmus paper for acidity before and after each experiment.

Porcelain evaporating dishes 120 mm. in diameter and 35 mm. deep were used as containers for the manure. Each was filled level full. The ammonium carbonate, when used, was imbedded in the manure; 57 grams was the amount generally employed. Ammonium hydroxide was not entirely satisfactory, because the ammonia escaped rapidly, and the addition of a liquid to the manure made it too wet. The controls held acidulated manure only, and were placed two, twenty-five, thirty or fifty feet from the acidulated manure which contained ammonium compounds. In one experiment the ammonium carbonate was placed in a glass dish to which water was added, and a dish containing acidulated manure was set at a distance of one foot on each side of it.

A summary of six experiments is given below. Each dish with its contents is spoken of as a lot:

Total egg-masses in 10 lots of HCl manure evolving ammonia from ammonium carbonate	164.0
Average per lot	16.4
Total egg-masses in 4 lots of HCl manure evolving ammonia from ammonium hydroxide	14.0
Average per lot	3.5
Total egg-masses in 10 lots of HCl manure separated 1–2 feet from ammoniated lots.	37.0
Average per lot	3.7
Total egg-masses in 10 lots of HCl manure separated 25, 30 and 50 feet from ammoniated lots	8.0
Average per lot	0.8

The lots which volatilized ammonia from ammonium carbonate were more than four times as attractive as the untreated acidulated lots placed near them (one to two feet), and more than twenty times as attractive as the acidulated lots placed some distance away (25 to 50 feet). In the single experiment in which an acidulated manure lot stood on each side of a dish containing ammonium carbonate and water, twelve egg-masses were deposited upon the acidulated manure, while none was found in the acidulated manure controls thirty feet distant. The oviposition response of the house fly in these experiments was roughly in an inverse ratio to the distance from the source of the ammonia.

Oviposition in Timothy Chaff and Pine Sawdust.—This series of experiments was conducted in the same manner as the acidulated manure series. The chaff and sawdust were always kept moist with water. The results are set forth in the following table:

Number	Material Used in Each Lot	Number of Lots	Distance from Ammoniated Lots	Duration of Experiments	Number of Egg-masses
1*a*	Timothy chaff and 241 gm. of ammonium carbonate	3		5 hours	19
b	Timothy chaff only	3	3 inches	5 hours	0
2*a*	Timothy chaff and 57 gm. of ammonium carbonate	1		17 hours	3
b	Timothy chaff only	1	2 feet	17 hours	0
c	Timothy chaff only	1	50 feet	17 hours	0
3*a*	Pine sawdust and 227 gm. of ammonium carbonate	3		99 hours	4
b	Pine sawdust only	3	12 inches	99 hours	0
4*a*	Pine sawdust and 106 gm. of ammonium carbonate	1		47 hours	2
b	Pine sawdust only	1	3 inches	47 hours	0

Timothy chaff which volatilized ammonia incited flies to oviposit on it within a short time. The average number of egg-masses per lot for the two experiments was 5.5, considerably lower than the average for the acidulated manure experiments. Larvæ were able to develop into normal flies in timothy chaff.

Pine sawdust was even less attractive than timothy chaff, with an average of 1.5 egg-masses per lot. Larvæ died soon after hatching in this substance.

Oviposition in Cotton and Filter Paper.—Pieces of ammonium carbonate were placed in evaporating dishes, covered with sterilized ab-

sorbent cotton, moistened with water, and exposed in a locality where flies were fairly abundant. Some of the dishes contained in addition small amounts of the following: ethyl alcoholic solution of skatol, ethyl alcoholic solution of indol, ethyl alcohol, phenol, valerianic acid, and butyric acid. In other dishes the ammonium carbonate was omitted, and the following compounds were added to the moistened cotton: ammonium sulphide solution, valerianic acid, and butyric acid. There were also controls of moistened cotton only.

For the filter paper experiments, the paper was torn into bits, moistened with water and placed over the ammonium carbonate. In one series the filter paper was stained with aqueous Bismarck brown.

Eleven experiments involving fifty-three individual lots showed positive results with only three combinations. These results are summarized below:

Material	Number of Experimental Dishes	Duration of Experiments	Number of Egg-masses Found
57 gm. ammonium carbonate + 2–5 c.c. valerianic acid + 50 c.c. water + cotton	7	3–72 hrs.	3
57 gm. ammonium carbonate + 2–5 c.c. butyric acid + 50 c.c. water + cotton	7	3–72 hrs.	18
57 gm. ammonium carbonate + 20–50 c.c. water + cotton	11	3–72 hrs.	1

Butyric acid, and to some extent, valerianic acid augmented the oviposition response of the house fly when added to moist ammoniated cotton. Ammonium carbonate and moist cotton without the aid of these acids brought forth almost no response.

Discussion

The small amount of oviposition in the distantly removed controls of the acidulated manure series was probably due to the fact that the flies were coaxed into the vicinity by the odor of ammonia from the ammoniated lots and came by chance to the distantly removed lots. These experiments show that many flies went a short distance from the exact source of the ammonia in order to place their eggs in a favorable substance and it is reasonable to expect a few would stray even farther. Of course a chemical substance present in the manure, but not tried in these experiments, may have been responsible for this slight attraction, or it may be true that an attractive odor is not always necessary to induce oviposition.

Female house flies have some power which enables them to discriminate between substances with high food value for their larvæ and substances which have little or no food value. This power is not infallible. Even when volatilizing ammonia, pine sawdust, cotton, or filter paper had little attraction, while acidulated horse manure and timothy chaff showed considerable attraction. It is suggested that this food-discriminating power is either a gustatory or a "contact-odor" perception.

Butyric and valerianic acids are found in barnyard manure, and it seems probable that their addition to ammoniated cotton gives to that substance an odor which simulates to a degree the odor arising from manure. If this is true it explains why house flies are readily attracted to ammoniated cotton to which these acids have been added. It is interesting to note that butyric and valerianic acids, when added in small amounts to ethyl alcohol, increased the attraction of the alcohol to *Drosophila ampelophila*.[5]

I hope to give these questions further attention. These studies emphasize the necessity for the proper disposal of all fermenting organic substances which volatilize ammonia, and reveal possible new angles of attack in the control of the house fly.

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[5] Barrows, William Morton, "The Reactions of the Pomace Fly, *Drosophila ampelophila* Loew. to Odorous Substances," *Jour. Exper. Zool.*, Vol. 4, pp. 515–537 (5 figs.).